

ABSTRACT OF THE DISCLOSURE

1 Quantitative and qualitative analysis of a nucleic acid analyte in a sample
2 suspected to contain the nucleic acid analyte is achieved by first preparing a reaction
3 mixture containing the sample and a known amount of an internal quantitation standard. At
4 least a first aliquot of the reaction mixture is combined with a set of amplification reagents
5 effective to amplify nucleic acids in the reaction mixture. The set of reagents includes at
6 least one primer pair which is effective to amplify a first region of the nucleic acid analyte
7 if present in the sample to produce a first amplified sample fragment and to amplify at least
8 a portion of the internal quantitation standard to produce a control fragment. Amplification
9 results in the formation of an amplification product mixture containing first amplified
10 sample fragments and control fragments when the nucleic acid analyte is present in the
11 sample, and only control fragments when the nucleic acid analyte is not present in the
12 sample. The relative amounts of first amplified sample fragments and control fragments
13 are analyzed to quantify the amount of nucleic acid analyte in the sample, and the sequence
14 of the first amplified sample fragments is determined to assess the qualitative
15 characteristics of any nucleic acid analyte. The internal quantification fragment is derived
16 from the analyte nucleic acid by the incorporation of a plurality of sequence variations.
17 These sequence variations include at least a first sequence variation effective to render the
18 internal quantitation standard distinguishable from the first amplified sample fragment, and
19 a second sequence variation effective to substantially eliminate the production of
20 sequencing products from interaction of the internal quantitation standard and the first
21 sequencing primer.